REMARKS

Reconsideration of the present application in view of the above amendments and following remarks is respectfully requested. As set forth above, Applicants have canceled claims 3, 5-11, and 14 without prejudice to the filing of any divisional, continuation, or, continuation-in-part application. Applicants hereby submit new claims 29-51. Applicants hereby note that original claim 5 has essentially been redrafted in independent form as claim 29. Support for new claims may be found in the subject application as originally filed, in part, at page 26, line 27 through page 27, line 24, and Figures 6 and 8 (*see, e.g.*, claim 29); at page 27, lines 18-22 (*see, e.g.*, claim 30); at page 23, lines 3-12 (*see, e.g.*, claim 31); at page 21, lines 17-20 (*see, e.g.*, claim 32); at page 5, lines 7-12 (*see, e.g.*, claims 33-36); at page 5, line 28 through page 6, line 3 (*see, e.g.*, claims 37-40); at page 5, lines 1-5 (*see, e.g.*, claims 41-42); at page 26, lines 8-13 (*see, e.g.*, claims 43-44); at page 21, line 29 through page 22, line 6 (*see, e.g.*, claim 46). Claims 1, 4, 15, and 16 have been hereby amended to more clearly define the subject matter encompassed by Applicants' invention, and claim 20 has been amended for mere editorial purposes to correct an inadvertent typographical error. Support for amended claim 4 may be found in the specification, in part, at page 22, lines 6-16. No new matter has been added. Therefore, claims 1, 2, 4, 12, 13, 15-18, 20, and 29-53 are currently pending.

Applicants respectfully submit herewith a Declaration by the Applicants, together with an Exhibit, pursuant to 37 C.F.R. §1.131. Also attached hereto is a marked-up version of the changes made to the specification and claims by the current amendment. The first page of the attached pages is captioned "Version With Markings to Show Changes Made."

OBJECTIONS

In the Office Action dated July 31, 2001, the Examiner has objected to claims 5, 10, and 14 for typographical errors (*i.e.*, omitted words). Applicants respectfully submit that this objection is now moot because claims 5, 10, and 14 have been canceled without prejudice, as set forth above.

REJECTION UNDER 35 U.S.C. §112, SECOND PARAGRAPH

In the Office Action, claims 5-7, 10, 11, and 14 were rejected under 35 U.S.C. §112, second paragraph, as indefinite. Applicants respectfully submit that this objection is now moot because claims 5-7, 10, and 14 have been canceled without prejudice, as set forth above. However, Applicants note that original claim 5 has essentially been redrafted in independent form as claim 29. Accordingly, Applicants respectfully submit that the invention as presently claimed satisfies the requirements of 35 U.S.C. §112, second paragraph.

REJECTIONS UNDER 35 U.S.C. §112, FIRST PARAGRAPH

(1) In the Office Action, claims 1-4, 8, 9, 12, 13, 15-18 and 20 were rejected under 35 U.S.C. §112, first paragraph, for lack of enablement. In particular, it is alleged that, in view of the state of the prior art and the instant specification, expression cassettes having more than four copies of a cationic peptide encoding sequence could not be successfully expressed in the absence of an anionic spacer.

Applicants submit that this rejection is now moot because claim 1 has been amended to more clearly define the subject matter encompassed by the instant invention. As presently claimed, the instant invention is directed, in pertinent part for this rejection, to a multi-domain fusion protein expression cassette comprising a promoter operably linked to a nucleic acid molecule that is expressed as an insoluble protein, wherein the nucleic acid molecule encodes a polypeptide comprising the structure (cationic peptide)-[(cleavage site)-(cationic peptide)]$_n$, wherein n is an integer having a value between one and four and at least one cationic peptide has antimicrobial activity. The instant specification concededly provides ample guidance to a person having ordinary skill in the art how to make and use a multi-domain fusion protein expression cassette according to the instant invention. Accordingly, Applicants respectfully submit that the claims satisfy the requirements of 35 U.S.C. §112, first paragraph and, therefore, request that this rejection be withdrawn.

(2) In the Office Action, claims 5-7, 10, 11, and 14 were rejected under 35 U.S.C. §112, first paragraph, for lack of enablement. In particular, it is alleged that the claimed expression cassettes are infinitely broad with regard to the number and position of the

cassette components. Therefore, it is asserted that in light of the prior art and guidance in the instant specification, a person having ordinary skill in the art would be required to experiment unduly to successfully make and use the full scope of the claimed invention.

As set forth above, Applicants respectfully submit that this objection is now moot because claims 5-7, 10, and 14 have been canceled without prejudice. However, as also set forth above, Applicants note that original claim 5 has essentially been redrafted in independent form as claim 29. Therefore, the instant invention is directed, in pertinent part for this rejection, to a multi-domain fusion protein expression cassette comprising a promoter operably linked to a nucleic acid molecule that is expressed as an insoluble protein, wherein the nucleic acid molecule encodes a fusion protein comprising (a) a carrier amino acid sequence, (b) an anionic spacer peptide, (c) at least two cationic peptides wherein at least one cationic peptide has antimicrobial activity, and (d) at least two cleavage sites wherein at least one cleavage site is between the cationic peptide and the carrier and at least one cleavage site is between the cationic peptide and the spacer, wherein the encoded fusion protein comprises the structure (carrier amino acid sequence)-[(cationic peptide)-(anionic spacer peptide)]$_n$-(cationic peptide) with n being an integer having a value between 1 and 100. Additionally, Applicants respectfully submit that the mere fact that exemplified embodiments in the specification are more limited than those recited in the claims does not provide sufficient reason for the Examiner to hold the claims as non-enabled. Applicants are not required to specifically exemplify all embodiments of the invention that are encompassed by the claims. The requirements of 35 U.S.C. §112, first paragraph can be fulfilled by the use of *illustrative* examples or by broad terminology. *In re Anderson*, 176 USPQ 331 (CCPA 1973).

Applicants respectfully submit that the disclosure of the instant specification is commensurate in scope with the claims and that no undue experimentation is required to practice the instant invention. Accordingly, Applicants respectfully submit that the claims satisfy the requirements of 35 U.S.C. §112, first paragraph and, therefore, request that this rejection be withdrawn.

REJECTION UNDER U.S.C. §102(a)

In the Office Action, claims 5-7, 10 and 14 were rejected under 35 U.S.C. §102(a) as anticipated by Zhang *et al. (Biochem. Biophys Res. Comm. 247*:674-680, 1998). In particular, it is asserted that Zhang *et al.* provide a multi-domain fusion protein expression cassette that expresses a fusion protein in insoluble form, as claimed. Applicants respectfully submit that this objection is now moot because claims 5-7, 10, and 14 have been canceled without prejudice, as set forth above. Furthermore, Applicants submit herewith a Declaration with an Exhibit providing evidence that Applicants have reviewed laboratory records and readily conclude that compositions of matter and methods as claimed in the subject application were conceived prior to 1998. Accordingly, in view of the pending claims and the foregoing remarks, Applicants respectfully submit that the rejections under 35 U.S.C. §102(a) have been overcome and, therefore, request that this rejection be withdrawn.

All of the pending claims in the application are now clearly allowable. Favorable consideration and a Notice of Allowance are earnestly solicited. The Examiner is urged to contact the undersigned attorney if there are any questions prior to allowance of this matter.

Respectfully submitted,

Seed Intellectual Property Law Group PLLC

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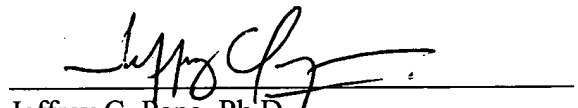
PATENT TRADEMARK OFFICE



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Enclosure:

Two Declarations under 37 C.F.R. §1.131
Exhibit

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VERSION WITH MARKINGS TO SHOW CHANGES MADE

In the Claims:

Claim 3, 5-11, and 14 have been canceled without prejudice.

Claims 1, 4, 15, 16, and 20 have been amended as follows:

1. (Amended) A multi-domain fusion protein expression cassette, comprising a promoter operably linked to a nucleic acid molecule which is expressed as an insoluble protein, wherein said nucleic acid molecule encodes a polypeptide comprising the structure (cationic peptide)-[(cleavage site)-(cationic peptide)]$_n$, wherein n is an integer having a value between ~~1 and 100~~ one and four and ~~said~~ at least one cationic peptide has antimicrobial activity.

2. The expression cassette according to claim 1 wherein said nucleic acid molecule also encodes a carrier protein.

4. (Amended) The expression cassette according to ~~claim 1~~ any one of claims 1 or 2 wherein said cleavage site can be cleaved by low pH or by a reagent selected from the group consisting of cyanogen bromide, N-chlorosuccinimide, 2-(2-nitrophenylsulphenyl)-3-methyl-3'-bromoindolenine, hydroxylamine, *o*-iodosobenzoic acid, Factor Xa, Factor XIIa, thrombin, enterokinase, collagenase, *Staphylococcus aureus* V8 protease, endoproteinase Arg-C, endoproteinase Glu-C, endoproteinase Lys-C, and trypsin.

12. The expression cassette according to claim 2 wherein said carrier protein is less than 100 amino acid residues in length.

13. The expression cassette according to claim 2 wherein said carrier protein is a truncated cellulose binding domain of less than 100 amino acids.

15. (Amended) The expression cassette according to ~~claim 1~~ any one of claims 1 or 2 wherein said promoter is selected from the group consisting of *lacP* promoter, *tacP* promoter, *trcP* promoter, *srpP* promoter, SP6 promoter, T7 promoter, *araP* promoter, *trpP* promoter, and λ promoter.

16. (Amended) A recombinant host cell comprising the expression cassette according to any one of claims ~~claim~~ 1, 2, 12, or 13.

17. The recombinant host cell of claim 16 wherein said host cell is a yeast, fungi, bacterial or plant cell.

18. The recombinant host cell of claim 17 wherein said bacterial host cell is *Escherichia coli*.

20. (Amended) A method of producing fusion proteins that contain a cationic peptide, comprising: ~~(a)~~ culturing the recombinant host cell of claim ~~15~~ 16 under conditions and for a time sufficient to produce said fusion protein.

Claims 29-53 have been added as follows:

29. (New) A multi-domain fusion protein expression cassette, comprising a promoter operably linked to a nucleic acid molecule that is expressed as an insoluble protein, wherein the nucleic acid molecule encodes a fusion protein comprising (a) a carrier amino acid sequence, (b) an anionic spacer peptide, (c) at least two cationic peptides wherein at least one cationic peptide has antimicrobial activity, and (d) at least two cleavage sites wherein at least one cleavage site is between the cationic peptide and the carrier and at least one cleavage site is between the cationic peptide and the spacer, wherein the encoded fusion protein comprises the structure (carrier amino acid sequence)-[(cationic peptide)-(anionic spacer peptide)]$_n$-(cationic peptide) with n being an integer having a value between 1 and 100.

30. (New) The expression cassette according to claim 29 further comprising at least one additional C-terminal anionic spacer peptide.

31. (New) The expression cassette according to claim 29 wherein the promoter is selected from the group consisting of *lacP* promoter, *tacP* promoter, *trcP* promoter, *srpP* promoter, SP6 promoter, T7 promoter, *araP* promoter, *trpP* promoter, and λ promoter.

32. (New) The expression cassette according to claim 29 wherein the carrier is selected from cellulose binding domain, glutathione-S-transferase, outer membrane protein F, β-galactosidase, protein A, or IgG-binding domain.

33. (New) The expression cassette of claim 29 wherein the carrier is located at the N-terminus of the fusion protein.

34. (New) The expression cassette of claim 29 wherein the carrier is located at the C-terminus of the fusion protein.

35. (New) The expression cassette according to claim 29 wherein the carrier is less than 100 amino acid residues in length.

36. (New) The expression cassette according to claim 35 wherein the carrier is a truncated cellulose binding domain.

37. (New) The expression cassette according to claim 29 wherein the anionic spacer has no cysteine residue.

38. (New) The expression cassette according to claim 29 wherein the number of anionic spacer peptides is greater than or the same as the number of cationic peptides.

39. (New) The expression cassette according to claim 29 wherein the number of anionic spacer peptides is less than the number of cationic peptides.

40. (New) The expression cassette according to claim 29 wherein the cumulative charge of the anionic spacer peptide reduces the cumulative charge of the cationic peptide.

41. (New) The expression cassette according to claim 29 wherein the fusion protein comprises from 2 to 40 cationic peptides.

42. (New) The expression cassette according to claim 29 wherein the fusion protein comprises from 2 to 20 cationic peptides.

43. (New) The expression cassette according to claim 29 wherein the cationic peptide is an indolicidin or analog thereof.

44. (New) The expression cassette according to claim 43 wherein the indolicidin or analog thereof is an indolicidin analog of up to 35 amino acids that comprises the sequence of I L K K W P W W P W R R K or I L R W P W W P W R R K.

45. (New) The expression cassette according to claim 29 wherein the cleavage site can be cleaved by low pH or by a reagent selected from cyanogen bromide, N-chlorosuccinimide, 2-(2-nitrophenylsulphenyl)-3-methyl-3'-bromoindolenine, hydroxylamine, *o*-iodosobenzoic acid, Factor Xa, Factor XIIa, thrombin, enterokinase, collagenase, *Staphylococcus aureus* V8 protease, endoproteinase Glu-C, endoproteinase Arg-C, endoproteinase Lys-C, chymotrypsin, trypsin, or a combination thereof.

46. (New) The expression cassette according to claim 29 wherein the cleavage site is with the carrier amino acid sequence, anionic spacer peptide, cationic peptide, or a combination thereof.

47. (New) A recombinant host cell comprising the expression cassette according to any one of claims 29-46.

48. (New) The recombinant host cell of claim 47 wherein the host cell is a yeast, a fungus, a bacteria or a plant cell.

49. (New) The recombinant host cell of claim 48 wherein the bacteria is *Escherichia coli.*

50. (New) A method of producing fusion proteins that contain a cationic peptide, comprising culturing the recombinant host cell of claim 47 under conditions and for a time sufficient to produce the fusion protein.

51. (New) The expression cassette according to any one of claims 1, 2, 29, or 30 wherein the expression cassette is contained in a vector or an expression vector.

52. (New) The recombinant host cell of claim 16 wherein the expression cassette is contained in an expression vector.

53. (New) The recombinant host cell of claim 47 wherein the expression cassette is contained in an expression vector.

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